

They have inflated anthers, which probably have a bellows-like action like the long stamen of *Solanum rostratum*⁷ and the anthers of *Rhexia Virginica*.⁸ Between the style and a long stamen is another long stamen with an anther like those of the short stamens. Bees, no doubt, force the pollen out of this as they do from the short stamens. The style is turned sometimes to the right, sometimes to the left, and the flower itself is turned slightly to one side or the other, so that the stigma touches the side of the visitor, making the flower *pleurotribe*. According to Meehan, the flowers fail to produce seed under a net. Both he and Leggett⁸ saw bumble-bees collecting the pollen. I have seen the flower visited for pollen by *Bombus americanorum* F. ♂.

Extranuptial nectaries.—Visitors: (on one occasion) A large red ant; *Sarcophaga* sp.; *Anthomyia* sp.; *Camptoneura picta* F. (Ortalidæ); *Coccinella sanguinea* L.
Carlinville, Ill.

Fermentation of bread.¹

KATHERINE E. GOLDEN.

Ferments have been known since very early times, for we have accounts of the early Egyptians using leaven to increase the lightness of bread. Much has been written and said in a superficial way about the fermentation of bread, and there are many methods of preparing and preserving leaven for bread-making given in old books, but what was in the leaven that produced the fermentation long remained an unsolved problem. Then came the early researches into the subject which established the now well-known facts that yeast causes carbon-dioxide and alcohol to be generated from sugar, that the carbon-dioxide causes the bread to rise, and that the alcohol is driven out of the bread by the heat in baking. The processes that the yeast and sugar underwent in causing the decomposition of the latter were not at that time understood, nor whether there were other organisms besides the yeast present.

Of late years, however, since bacteriology has received the attention of scientific men, the old view that yeast alone

⁷Todd: l. c. ⁸Torr. Bull. V III, 102-104.

¹Part of a thesis presented to the faculty of Purdue University for the bachelor's degree, based upon work done in the botanical laboratory under the direction of Dr. J. C. Arthur.

causes bread to rise has been questioned somewhat, some still claiming that it does, others that it has nothing whatever to do with the rising, while still others take a halfway course, that is, that yeast and bacteria acting together do the work.

Chicandard,² in 1883, presented a paper before the French Academy of Sciences in which he explained the fermentation of bread to be the result, not of yeast, but of a special bacillus that develops normally in the dough, while the yeast only hastens the development. He claims that the most essential part of the fermentation is the transformation of a part of the insoluble albuminoids into soluble ones.

Laurent,³ writing four years later, presents the same idea in regard to the cause of the fermentation. He found in dough a bacterium, *Bacillus panificans*, as he calls it, that occurs in short and long rods and forms sharply defined yellow colonies on plate-cultures. It can stand a high degree of heat, so high that the rods are said to be still alive under the crust of the bread. The spores will stand long heating at boiling temperature. Laurent says that this bacillus is responsible for the formation of carbon-dioxide, besides a certain amount of lactic, butyric and acetic acids. In the warm season this bacillus sometimes causes the bread to become slimy, so that it can be drawn out into threads. In such bread are found myriads of the organisms which change the starch into erythrodextrin, and thus bring about the slimy metamorphosis.

Wigand⁴ agrees in substance with the preceding views in regard to bread fermentation, but he says that the bacillus is formed spontaneously from the albumen of the gluten, for although an eminent scientist and writing but six years ago, he believed in the now generally discarded theory of spontaneous generation.

And lastly, Marcano⁵ believes that the motile bacteria found in dough are the true cause of the fermentation.

Opposed to the view that bacteria are the cause of the rising of bread, we have the opinions of such men as Birnbaum, Arcangeli and Dünnerberger. Birnbaum⁶ thinks that

²Chicandard, Comptes rend., 1883, p. 1585; quoted by Dünnerberger, l. c. and Peters l. c.

³Laurent, E. Bull. Acad. Roy. Sci. Belg., Vol. X, 1885, p. 765; Bull. Soc. Roy. Bot. Belg., Vol. XXV., pt. 2 p. 165; abstract in Centralb. f. Bak. u. Parasitenk., Vol. I, 1887, p. 504; Just's Bot. Jahresb., Vol. XIV, pt. 1, 1886, pp. 396, 397.

⁴Wigand, Entstehung und Fermentwirkung der Bakterien, 2d Ed., 1884, p. 11.

⁵Marcano, Compt. rend., xvi, xvii, 1883; quoted by Dünnerberger, l. c.

⁶Birnbaum, Lehrbuch der landwirtschaftlichen Gewerbe, 1866, Vol. I, p. 228; quoted by Dünnerberger, l. c.

the action of leaven is due solely to the presence in it of common or alcoholic yeast. Arcangeli⁷ also thinks yeast is the cause of the fermentation. In every instance he found a bacterium, the common *Bacillus subtilis*, but he says this is of very little concern except in facilitating the solution of the albuminous bodies. Dünnerberger⁸ goes even farther than this, for he claims the bacteria found in bread are a pollution and entirely dispensable.

Then come the views of Boutroux, Flügge and Peters which disagree with those just stated, that is, that yeast alone or bacteria alone are the cause of the rising of the bread. They found other organisms besides the yeast in the dough, and they claim that these organisms assist in the rising. Boutroux⁹ thinks that both yeast and bacteria assist in the fermentation, while Flügge¹⁰ thinks that the bacteria may help in the fermentation since they are found in leaven in overwhelming quantities. Peters¹¹ has studied leaven more particularly. He found five different bacteria in it that had more or less resemblance to one another. He holds that common yeast causes the alcoholic fermentation and bacteria the acid fermentation. But he thinks the bacteria are of secondary importance.

There are many points of difference in the opinions just cited, and at first thought it seems rather unaccountable that men who worked with scientific precision, as these men undoubtedly did, should arrive at such different results. The differences can doubtless be accounted for, however, if we take into consideration the fact that they worked under different conditions and probably looked at the subject from different standpoints, for though it is presumable from their writings that nearly all of them used leaven, it was very probably obtained by somewhat different methods and under different conditions.

Leaven is dough left over from one baking to another, either with or without the addition of an extract of hops or malt. This is the kind of ferment that is generally used in the old countries. In this country in places distant from

⁷ Arcangeli, G. Atti. Soc. Toscano Sci. Nat., Vol. IX, 1888, p. 22; abstract in Centralb. f. Bak. u. Parasitenk., 1888, Vol. III, p. 717.

⁸ Dünnerberger, C. Bot. Centralb., Vol. XXXIII 1888, p. 245; Archiv der Pharmacie, Vol. XXVI, 1888, p. 546; abstract in Bot. GAZETTE, Vol. XIII, 1888, p. 140.

⁹ Boutroux, Compt. rend., XCVII., 1883; quoted by Dünnerberger, l. c.

¹⁰ Flügge, Die Mikroorganismen, 1886, p. 491.

¹¹ Peters, W. L. Die Organismen des Sauerteigs und ihre Bedeutung für die Brotgä-
rung, Inaug.-Diss., 1889; Bot. Zeit., XLVII., 1889, p. 405; abstract in Jour. Roy. Micr. Soc.,
1890, p. 75.

markets a ferment is made from potatoes, boiled and mashed, with flour, salt, sugar, the water that the potatoes have been boiled in, and yeast. There are various other methods of making this ferment, but they do not differ essentially from one another. This is sometimes called emptyings, or jug-yeast, and is semi-liquid, so that it differs very materially from the leaven of the old country. Of course, in towns and cities where a fresh supply of yeast can be obtained readily no such methods need be resorted to. Besides the ferments mentioned there are also the dry yeast cakes, that is, yeast mixed with corn-meal, and dried, which will vegetate when moistened, and the salt-rising where no ferment is added, the fermentation being supposed to be set up by the organisms that are already in the ingredients.

The experiments which I performed in order to determine whether the yeast or the bacteria are more instrumental in causing bread to rise do not solve the question by any means, still they give some additional information on the subject. Freshly made dough that had been fermented with Vienna pressed yeast, commonly called German yeast, and sold under the name of Fleischmann's Compressed Yeast, was examined with the microscope and the yeast found to be *Saccharomyces cerevisiæ*, and with it a bacterium having the characteristics of *Bacillus subtilis*. These two germs were separated from each other by means of gelatine plate-cultures. A single colony of each was placed in flasks in equal quantities of a nutrient solution made according to directions given by Dr. Stone,¹² the proportions being 25 grams German yeast to 125 cubic centimetres distilled water, with 10 per cent. sugar, boiled thoroughly, filtered, and sterilized for three successive days. The flasks containing the yeast and bacteria were then placed in a vegetation chamber, kept at about 84-86° F. (29-30° C.), this being the optimum temperature for *Bacillus subtilis*, that of yeast being about 92° F. (33° C.). After vegetating for two and one-half months they were tested for the amount of carbon-dioxide given off by each, the gas that had accumulated in the flasks being first drawn off. The yeast gave off 23 mg. gas in one hour, the bacteria 70 mg. in the same time. A second test was made of a four days' growth of each, the germs for these growths being obtained from agar cultures of each, inoculated from the original plate cultures. The yeast gave off 50 mg., the bacteria 60 mg. in an hour. In a case where

¹² Stone, W. E., BOT. GAZETTE, 1887, p. 270.

the sponges, made as will be shortly stated, and still in the flasks, were tested instead of the inoculated fluids for the carbon-dioxide, the yeast gave 90 mg. in an hour, the bacteria only 10 mg. in the same time.

After each test a bread sponge was made from both kinds, that is 200 grams sterilized flour was put into a flask and with the flour a nutrient solution, consisting of 150 c.c. potato-broth, 2 grams salt, and 8 grams sugar. The bread sponges were then placed in a vegetation chamber and kept at uniform temperature of 84-86° F. (29-30° C.), for about twenty-four hours. At the end of that time the yeast sponge had run well and uniformly, but the bacteria sponge invariably showed a separation of the flour from the liquid, the flour going to the bottom of the flask, and a layer of clear liquid remaining on the surface.

The sponges then had more flour added to them, and the dough thus formed was kneaded thoroughly, and again placed in the vegetation chamber to rise, about twenty-four hours being allowed for this; a second kneading was then done, the time allowed for rising being about the same as after the first kneading. The yeast dough rose higher than the other in every case, but in the kneading felt like bread with shortening in it, lacking the tough, elastic qualities of good dough. The bacteria dough, though not risen so high, felt more like the ordinary dough, being more elastic, tougher, and smelling somewhat sweeter, though it, too, felt as if it contained some shortening.

The dough when placed in the baking-pans was allowed to rise for about six hours before transferring to the oven. It was then baked at a temperature of 280-350° F. (138-176° C.).

The yeast bread in every case showed a greater degree of lightness, as indicated by its bulk, than the other, but was coarse in texture, being filled with numerous large cavities. The bacteria bread, though apparently not so well risen, had a finer texture, with but the occasional occurrence of large cavities.

The sterilized flour used in all the experiments was prepared by first drying it in an oven two or more hours a day for four to six days, the oven being kept at a temperature of 150-220° F. (66-105° C.). The oven was then raised to 250-300° F. (121-149° C.), for an hour and the sterilization completed.

The utensils used were in all cases sterilized by heat,

and the hands of the operator washed in corrosive sublimate, but perfectly pure cultures were not obtained in any instance.

Though the cultures were not perfectly pure, they had such a very small amount of impurities in them that the results were almost or quite the same as would very probably be obtained from pure cultures.

The inferences drawn from these experiments are that both yeast (*Saccharomyces cerevisiæ*) and bacteria (*Bacillus subtilis*?) separately generate carbon-dioxide in sufficient quantities to raise bread. The amount of gas generated is presumably in direct proportion to the growth of the organisms and the viscosity of the surrounding medium. From the fact that the bacteria-inoculated fluid gave off more carbon-dioxide than the yeast fluid, while the bacteria sponge gave off less than the yeast sponge, it would indicate that the growth of yeast was less in the fluid than the growth of bacteria, but greater in the sponge, as the sponges in both cases were most probably of equal viscosity, as the conditions were as nearly as possible the same in both.

It was demonstrated by the experiments that both yeast and bacteria can separately raise bread, and, under the conditions of the experiments, the yeast somewhat better than the bacteria. Now, whether or not they act together in raising bread ordinarily was not demonstrated, but from the fact that both organisms were found in large quantities in dough that had been raised by Fleischmann's yeast, and that bacteria are always in the air and in large quantities on the surface of the grain from which flour is made, and also that they occur in all preparations of yeast ferment, it seems to be the only satisfactory conclusion that both the bacteria and the yeast act together in raising most if not all kinds of bread.

Purdue University, Lafayette, Ind.

BRIEFER ARTICLES.

Some effects of the mild winter.—The mild weather during the past winter caused so many remarkable changes in the usual habits of plants, in this locality, that I can not forbear offering a few notes for publication. *Malva rotundifolia*, *Stellaria media*, *Alyssum maritimum*, *Capsella Bursa-pastoris*, *Bellis perennis*, and *Lamium amplexicaule*, continued in bloom all winter, except about the first three weeks of March, during which time it was so cold as to stop vegetable growth. The cold spell